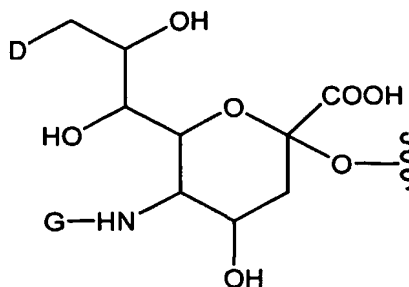


WHAT IS CLAIMED IS:

- 1 1. A Granulocyte Colony Stimulating Factor peptide comprising the moiety:



2

3

wherein

4

D is a member selected from -OH and R^1 -L-NH-;

5

G is a member selected from R^1 -L- and $-C(O)(C_1-C_6)alkyl$;

6

R^1 is a moiety comprising a member selected a moiety comprising a straight-chain or branched poly(ethylene glycol) residue; and

7

8

L is a linker which is a member selected from a bond, substituted or

9

unsubstituted alkyl and substituted or unsubstituted heteroalkyl,

10

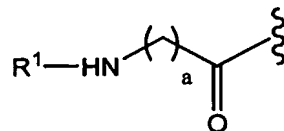
such that when D is OH, G is R^1 -L-, and when G is $-C(O)(C_1-C_6)alkyl$, D is

11

 R^1 -L-NH-.

1

2. The peptide according to claim 1, wherein L- R^1 has the formula:



2

3

wherein

4

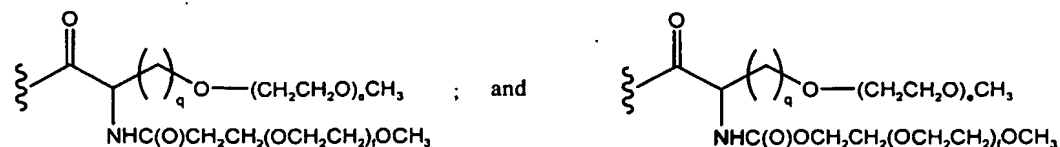
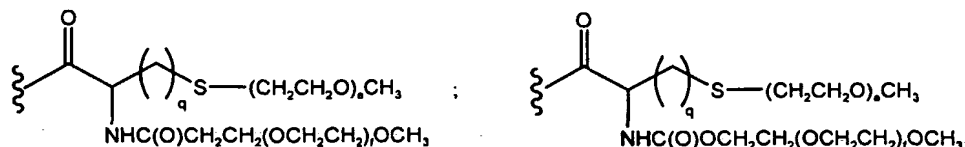
a is an integer from 0 to 20.

1

3. The peptide according to claim 1, wherein R^1 has a structure that is a member

2

selected from:



3

4

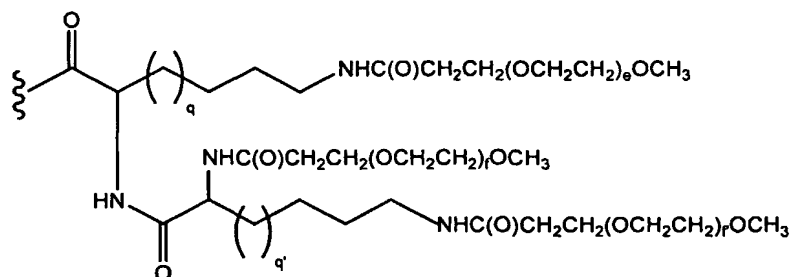
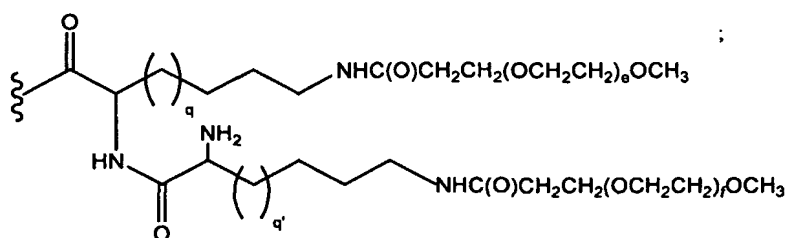
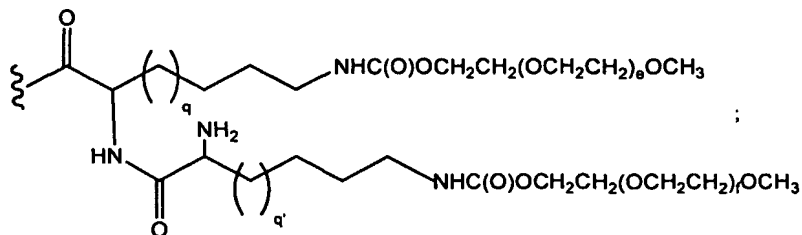
wherein

5 e and f are integers independently selected from 1 to 2500; and

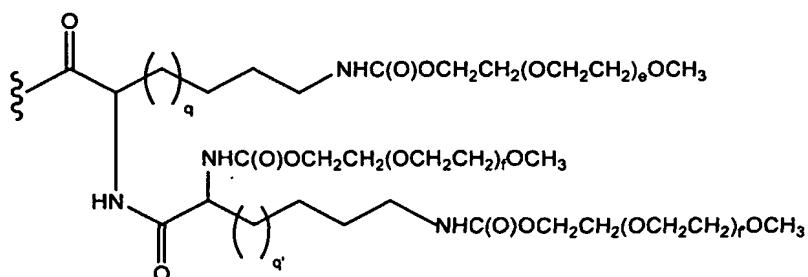
6 q is an integer from 0 to 20.

1 4. The peptide according to claim 1, wherein R¹ has a structure that is a member

2 selected from:



and

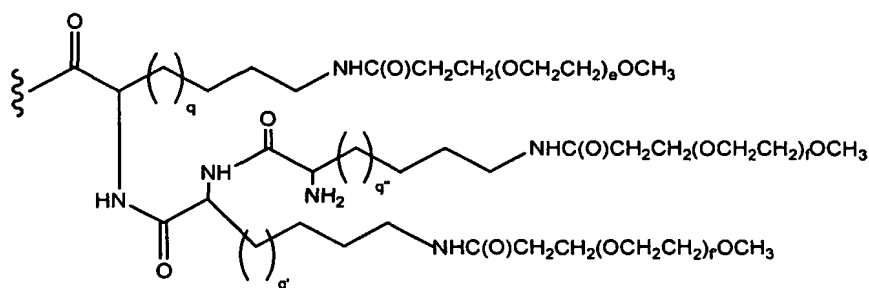
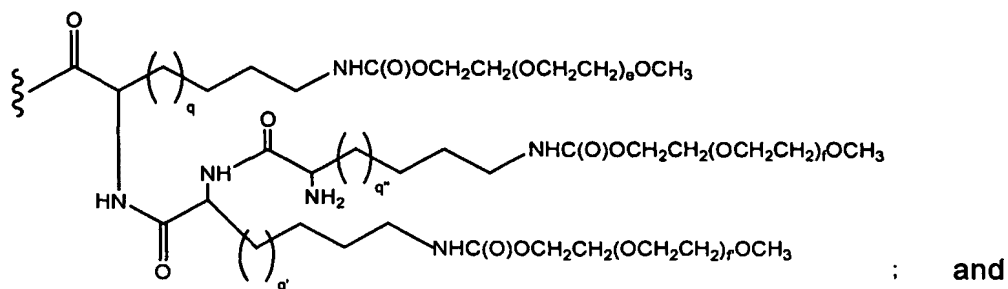


3 wherein

4 e, f and f' are integers independently selected from 1 to 2500; and

5 q and q' are integers independently selected from 1 to 20.

- 1 5. The peptide according to claim 1, wherein R^1 has a structure that is a member
2 selected from:



3

4

wherein

5

e, f and f' are integers independently selected from 1 to 2500; and

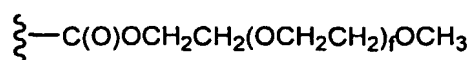
6

q, q' and q'' are integers independently selected from 1 to 20.

1

2

6. The peptide according to claim 1, wherein R^1 has a structure that is a member
selected from:



3

4

wherein

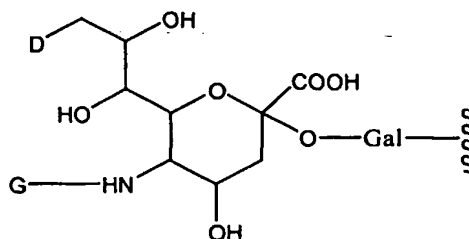
5

e and f are integers independently selected from 1 to 2500.

1

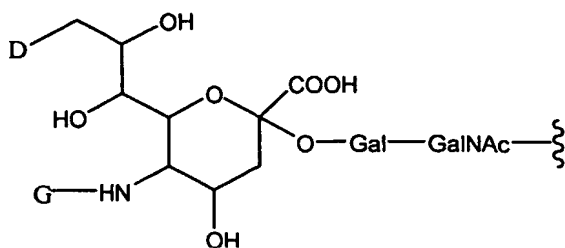
2

7. The G-CSF peptide according to claim 1, wherein said moiety has the
formula:

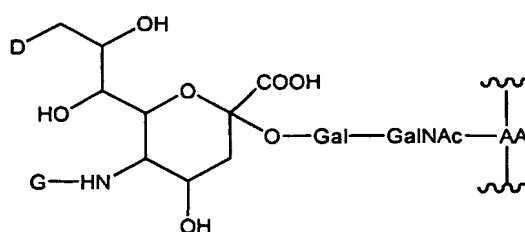


3

- 1 **8.** The G-CSF peptide according to claim 1, wherein said moiety has the
2 formula:



- 1 **9.** The G-CSF peptide according to claim 1, wherein said moiety has the
2 formula:



- 3
4 wherein

5 AA is an amino acid residue of said peptide.

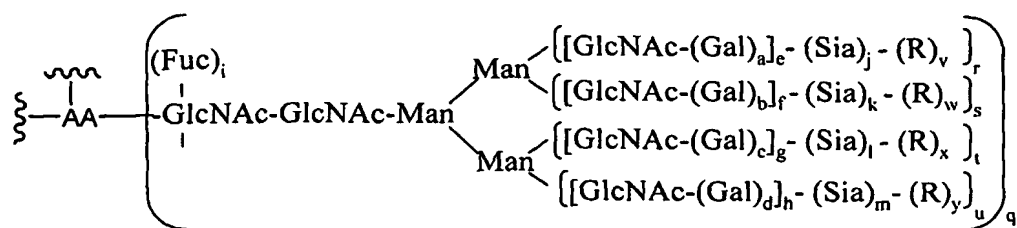
- 1 **10.** The G-CSF peptide according to claim 9, wherein said amino acid residue is a
2 member selected from serine or threonine.

- 1 **11.** The G-CSF peptide according to claim 1, wherein said peptide has the amino
2 acid sequence of SEQ. ID. NO:1.

- 1 **12.** The G-CSF peptide according to claim 11, wherein said amino acid residue is
2 threonine at position 133 of SEQ. ID. NO:1.

- 1 **13.** The peptide according to claim 1, wherein said peptide has an amino acid
2 sequence selected from SEQ. ID. NO:1 and SEQ ID NO:2.

- 1 **14.** The G-CSF peptide according to claim 1, wherein said moiety has the
2 formula:



3

4 wherein

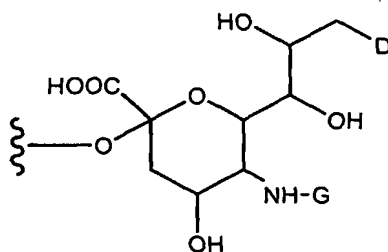
5 a, b, c, d, i, r, s, t, and u are integers independently selected from 0 and 1;

6 q is 1;

7 e, f, g, and h are members independently selected from the integers from 0 to
8 6;9 j, k, l, and m are members independently selected from the integers from 0 and
10 100;11 v, w, x, and y are independently selected from 0 and 1, and least one of v, w, x
12 and y is 1;

13 AA is an amino acid residue of said G-CSF peptide;

14 Sia-(R) has the formula:



15

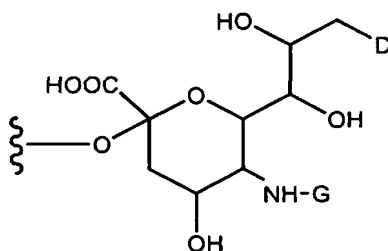
16 wherein

17 D is a member selected from -OH and R¹-L-NH-;18 G is a member selected from R¹-L- and -C(O)(C₁-C₆)alkyl;19 R¹ is a moiety comprising a member selected a straight-chain or
20 branched poly(ethylene glycol) residue; and21 L is a linker which is a member selected from a bond, substituted or
22 unsubstituted alkyl and substituted or unsubstituted heteroalkyl,
23 such that when D is OH, G is R¹-L-, and when G is -C(O)(C₁-C₆)alkyl,24 D is R¹-L-NH-.

1 15. The peptide according to claim 14, wherein said amino acid residue is an
2 asparagine residue.

1 16. The peptide according to claim 1, wherein said peptide is a bioactive
2 Granulocyte Colony Stimulating Factor peptide.

1 17. A method of making a G-CSF peptide conjugate comprising the moiety:

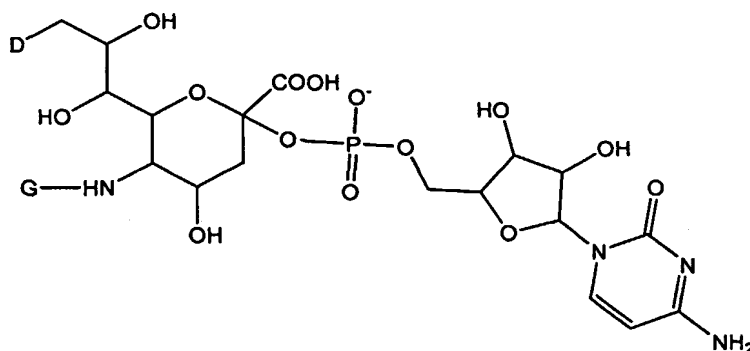


2
3 wherein

4 D is a member selected from -OH and R^1 -L-HN-;
5 G is a member selected from R^1 -L- and $-C(O)(C_1-C_6)alkyl$;
6 R^1 is a moiety comprising a member selected a straight-chain or branched
7 poly(ethylene glycol) residue; and
8 L is a linker which is a member selected from a bond, substituted or
9 unsubstituted alkyl and substituted or unsubstituted heteroalkyl,
10 such that when D is OH, G is R^1 -L-, and when G is $-C(O)(C_1-C_6)alkyl$, D is
11 R^1 -L-NH-,

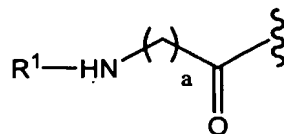
12 said method comprising:

13 (a) contacting a substrate G-CSF peptide with a PEG-sialic acid donor moiety
14 having the formula:



15
16 and an enzyme that transfers said PEG-sialic acid onto an amino acid
17 or glycosyl residue of said G-CSF peptide, under conditions
18 appropriate for the transfer.

- 1 **18.** The method according to claim 17, wherein L-R¹ has the formula:

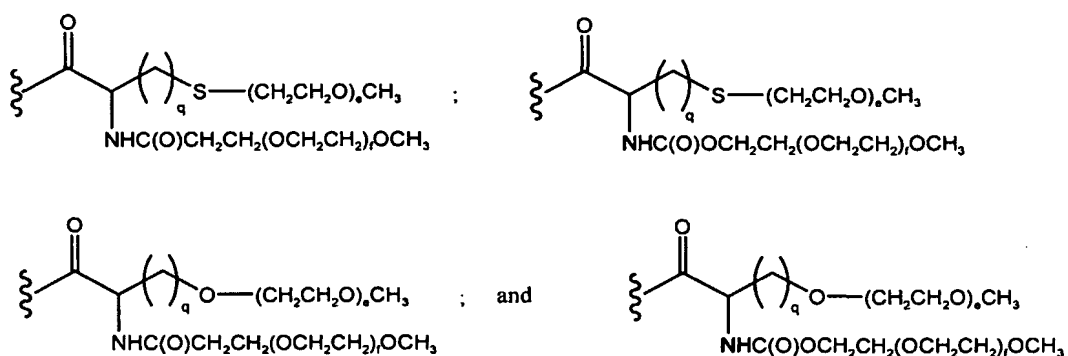


2

3 wherein

4 a is an integer from 0 to 20.

- 1 **19.** The method according to claim 17, wherein R¹ has a structure that is a
2 member selected from:



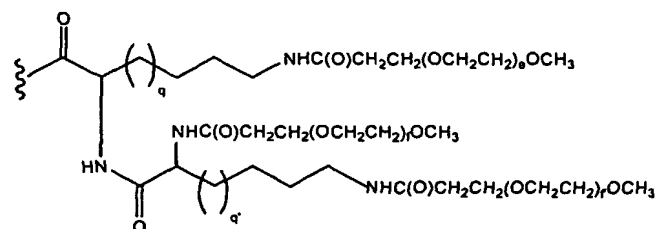
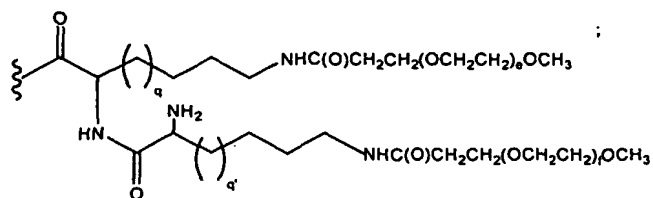
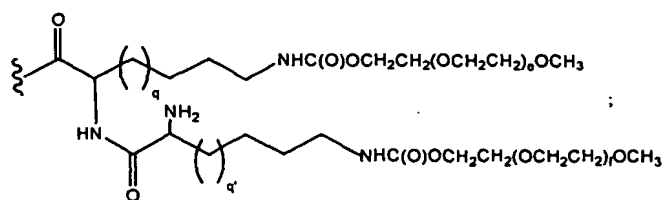
3

4 wherein

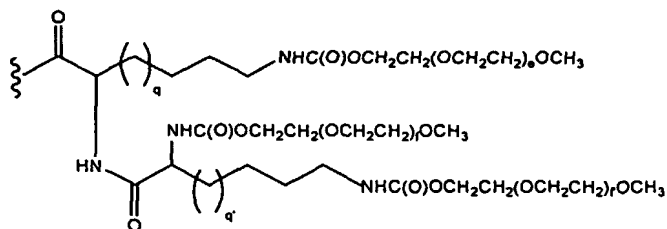
5 e and f are integers independently selected from 1 to 2500; and

6 q is an integer from 0 to 20.

- 1 **20.** The method according to claim 17, wherein R¹ has a structure that is a
2 member selected from:



; and



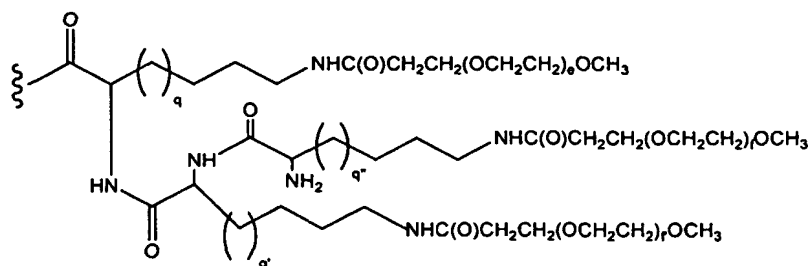
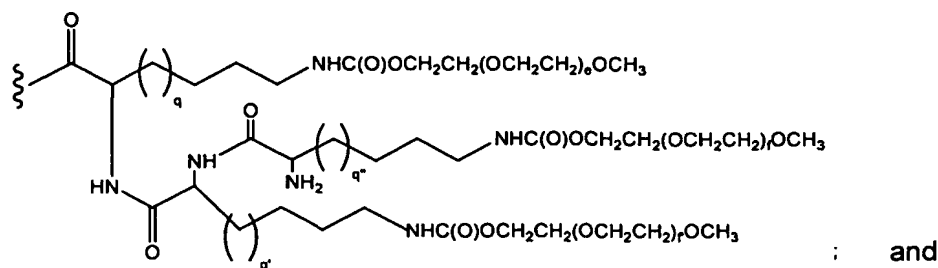
3

4 wherein

5 e, f and f' are integers independently selected from 1 to 2500; and

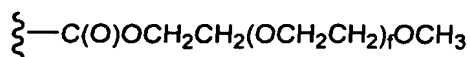
6 q and q' are integers independently selected from 1 to 20.

- 1 21. The method according to claim 17, wherein R^1 has a structure that is a
2 member selected from:



- 3
4 wherein
5 e, f and f' are integers independently selected from 1 to 2500; and
6 q, q' and q'' are integers independently selected from 1 to 20.

- 1 22. The method according to claim 17, wherein R^1 has a structure that is a
2 member selected from:



- 3
4 wherein
5 e and f are integers independently selected from 1 to 2500.

- 1 23. The method of claim 17, further comprising, prior to step (a):
2 (b) expressing said substrate Granulocyte Colony Stimulating Factor
3 peptide in a suitable host.

- 1 24. The method of claim 17, wherein said host is selected from an insect cell and a
2 mammalian cell.

- 1 25. A method of stimulating inflammatory leukocyte production in a mammal,
2 said method comprising administering to said mammal a peptide according to claim 1.

1 **26.** A method of treating infection in a subject in need thereof, said method
2 comprising the step of administering to the subject an amount of a peptide according
3 to claim 1, effective to ameliorate said condition in said subject.

1 **27.** A pharmaceutical formulation comprising the Granulocyte Colony Stimulating
2 Factor peptide according to claim 1, and a pharmaceutically acceptable carrier.

1 **28.** A method of refolding an insoluble recombinant granulocyte colony
2 stimulating factor (GCSF) protein, the method comprising the steps of:
3 (a) solubilizing the GCSF protein; and
4 (b) contacting the soluble GCSF protein with a buffer comprising a
5 redox couple to refold the GCSF protein, wherein the refolded GCSF protein is
6 biologically active.

1